Exotic crane flies *T. paludosa* and *Tipula oleracea* in Oregon

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There is great crane fly diversity in the PNW due to the moist environments. However, majority of the native species are saprophytic and not known as pests in cropping systems. Two exotic species, *Tipula paludosa* and *T. oleracea*, that are pests in their native environments in Europe, have been introduced inadvertently into Oregon. In Europe *T. paludosa* is known as a pest in grasslands and spring cereals. It was introduced to British Colombia in 1965 and first reported on the northwest coast in Oregon in 1989 as a pest in pastures in Tillamook County. *T. oleracea* is less dominant as a pest in Europe compared with *T. paludosa* but it is reported damaging winter cereals and crucifers. It was first reported in the PNW in 1998.

There is considerable sympatry in the distribution of *T. paludosa* and *T. oleracea* in the PNW, but they differ in their life cycles and feeding patterns. *T. paludosa* adults emerge in early fall, mate soon after emergence, and females lay eggs on the ground. Eggs hatch in September, and larvae feed voraciously in winter and spring. However, feeding is reduced at the end of spring, and the larvae undergo summer aestivation (Fig. 1). Pupation occurs in late summer. In contrast, *T. oleracea* has two generations a year. Adults emerge in March-April and again in September -October. There is no period of aestivation and larvae are present and active in the soil all year round. Based on this difference in life cycles, there is a variation in periods when damaging stages of both species are present in agricultural fields (Fig. 1). Damage by *T .oleracea* can occur throughout the year, while *T. paludosa* causes little additional damage in the summer. Hence different management strategies are needed for each species.

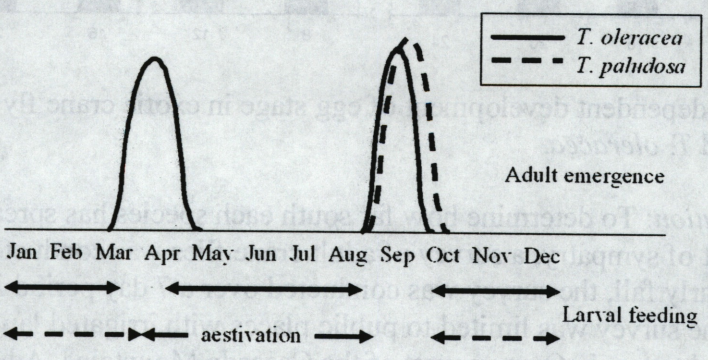


Fig. 1. Periods of adult emergence (above line) and larval feeding (below line).

Tipula oleracea and *T. paludosa* also differ in dispersal patterns. *T. paludosa* adult females are gravid at emergence and unable to travel any distance before ovipositing. This can lead to build up of local populations. *T. oleracea* females, on the other hand, are better fliers than *T. paludosa*, so eggs are more widely dispersed.

T. paludosa and *T. oleracea* are similar in appearance. They can be separated as adults based on eye and antennal characters. Larvae are dimorphic with regard to the anal papillae. However the reliability of this character has been questioned and currently, larvae need to be reared through to adults for identification. This is time consuming since *T. paludosa* takes a year for development. The trigger breaking summer aestivation is unknown, there is extensive mortality prior to pupation and few adults emerge.

To better understand the biology of the two species, we are studying temperature-dependent development and distribution of *T. paludosa* and *T. oleracea*. In addition we are developing a molecular tool for separation of the two species at the larval stage.

Temperature-dependent development: Field collected *T. paludosa* and *T. oleracea* were mated for collection of eggs. Eggs and first instar larvae were placed in incubators set at 0°, 4°, 8°, 12°, 16°, 24°, 28° and 32° C. The number of days to hatch at each temperature, and mortality at each temperature, are currently being recorded. The study is in progress, but preliminary data indicates that *T. oleracea* development is more rapid at a range of temperatures (8° through 24°C) (Fig. 2). Data on lower and upper development thresholds for each species, which will be generated in this study, will be used for predictions on range expansion of the two exotic species.

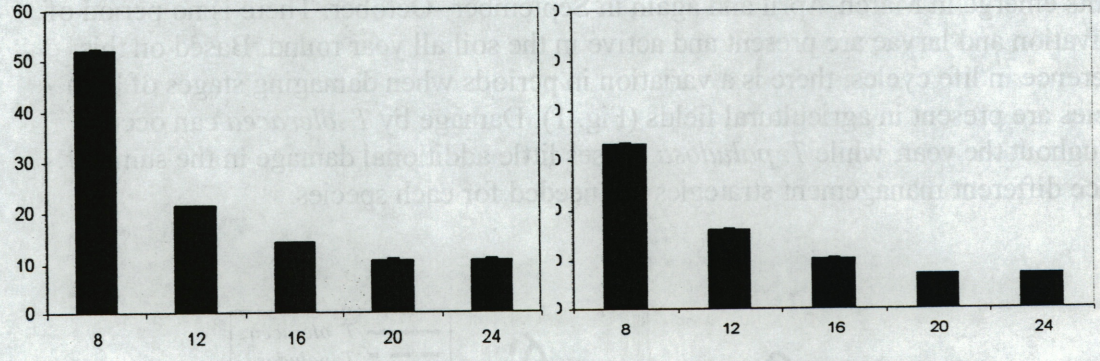
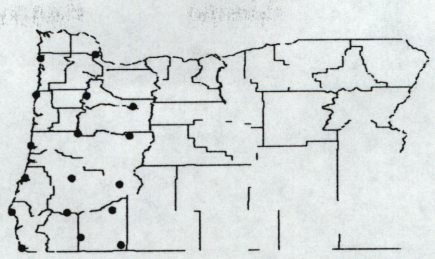
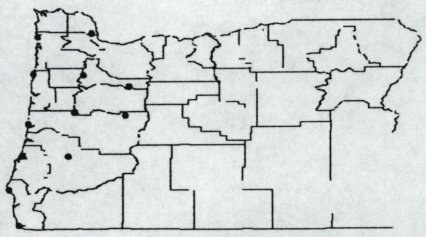


Fig.2. Temperature-dependent development of egg stage in exotic crane fly species *Tipula paludosa* and *T. oleracea.*

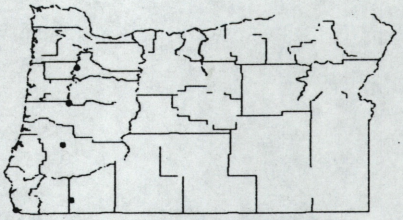
Geographic distribution: To determine how far south each species has spread, and to determine the extent of sympatry, a survey of adult crane flies was conducted. Since both species emerge in early fall, the survey was conducted over a 7 day period in mid September 2003. The survey was limited to public places with irrigated lawns such as parks in incorporated towns in Oregon west of the Cascade Mountains. Adults were caught, transported to the laboratory and identified. Out of 17 sites sampled, *T. paludosa* was collected from 12 sites while *T. oleracea* was collected from 5 (Fig. 3). At 4 sites, both species were collected including a site on the coast at the border of OR and CA. However, at the inland site close to the CA border, only *T. oleracea* was collected. In addition, only *T. paludosa* was collected at the 2 eastern sites at the foothills of the Cascades.



All sites sampled



T. paludosa



T. oleracea

Fig. 3. Distribution of *T. paludosa* and *T. oleracea* based on adult survey conducted west of Cascades in September 2003.

Molecular Marker: DNA was extracted from single third or fourth instar larvae of *Tipula oleracea*, *T. paludosa,* and an unknown larva collected from a golf course in Portland. From each individual larva, mitochondria *cytB* was amplified with primers from the mosquito mitochondrial genome. A single, ca. 400 bp band, was obtained for each sample. Amplicons were purified and sequenced. The resulting sequences were compared to the public DNA sequence database Genbank, and confirmed as Diptera sequences. Sequences were 410 bp long (excluding primer sequences) and the two known samples had 7.3% sequence divergence. The unknown sample was identical to *T. oleracea*. In subsequent analyses, we recorded 21 % and 27 % sequence divergence between a native species and *T. oleracea* and *T. paludosa*, respectively.

The molecular marker has several advantages over traditional rearing (Table 1 and was used for identification of larvae collected from diverse cropping systems in the Willamette Valley in Oregon. Twenty samples of *T. paludosa* were collected from lawns and peppermint fields while *T. oleracea* was collected from turf, lawns, peppermint, annual and perennial rye grass fields.

Table 1. Comparison of exotic *Tipula* spp. identification using molecular marker with traditional rearing

Identification	Molecular Analysis	Rearing
Time	2-3 days	up to 1 year for *T. paludosa*
Cost	$ 15-20	$ 416 (primarily for labor)
Mortality	none	extensive for *T. paludosa* (aestivation trigger still unknown)
Preservation	easy - in alcohol; freezer	not applicable